Comments on the Search for the Source of the Genetic Code: A Correction

Brian K. Davis

Research Foundation of Southern California, Inc.,
P.O. Box 13595, La Jolla CA 92038, U.S.A.

In evaluating the '2x2 code' scenario [1] for the origin of the genetic code, twenty-five pairs of tRNA attributed to the Last Common Ancestor, [2] with complementary anticodon triplets, were inspected for 2-base and dual complementarity. A tRNA in three pairs (8, 16, 24) was inadvertently given an incorrect acceptor-stem second base. [3] The results of an updated statistical analysis are presented here. They confirm my previous conclusion: early tRNA species do not display either form of complementarity.

There are eight (8/12) ancestral tRNA pairs with complementary second bases in the 3'-NUN/5'-NAN set and eight (8/13) in the 3'-NGN/5'-NCN set (Table 1). A semi-match occurs in seven pairs (1, 4, 15, 19, 20, 23, 24), with 2-base identity resolved only to a G or C, reducing the total number of complementary pairs from 16 to 12.5. 2-Base complementarity among the twenty-five tRNA pairs examined is non-significant, $p(x \geq 12.5) = 0.65$ NS, with a better than one-in-two chance of occurring randomly. The frequency of complementary pairs remains non-significant, $p(x \geq 16) = 0.12$ NS, even when each semi-matched pair is treated as a full-match. [3]

Table 1. Frequency of complementary second bases in acceptor stem of pairs of amino acid adaptors with complementary anticodons[1]

Amino[2] Acids	second base TRNA Stem	anti-codon	Compl.	Dual Compl.[3]	Amino[2] Acids	Second base tRNA stem	Anti-Codon	compl.	dual compl.
	3'-NUN/5'-NAN					3'-NGN/5'-NCN			
1. Asp/ Val	**C** C,**G**	CUG/ GAC	½+	-	14. Ala/ Gly	**G** **C**	C**G**G/ G**C**C	+	+
2. Glu/ Phe	C C	CUU/ GAA	-	-	15. Ala/ Cys	**G** **C**,G	C**G**U/ G**C**A	½+	½+
3. Glu/ Leu	C C	CUC/ GAG	-	-	16. Ala/ Arg	G G	CGC/ GCG	-	-
4. Asn/ Val	**C** C,**G**	UUG/ GAC	½+	-	17. Thr/ Gly	C C	UGG/ GCC	-	-
5. Lys/ Phe	N C	UUU/ GAA	-	-	18. Thr/ Ser	C N	UGA/ GCU	-	-
6. Lys/ Leu	C C	UUC/ GAG	-	-	19. Thr/ Cys	**C** C,**G**	UGU/ GCA	½+	-
7. His/ Val	**C** **G**	GUG/ CAC	+	-	20. Thr/ Arg	**C** **G**,C	UGC/ ACG	½+	-
8. Gln/ Leu	G C	GUU/ CAA	+	-	21. Pro/ Gly	**G** **C**	G**G**G/ C**C**C	+	+
9. Gln/ Leu	**G** **C**	GUC/ CAG	+	-	22. Pro/ Trp	G G	GGU/ CCA	-	-
10 Tyr/ Val	**C** **G**	AUG/ UAC	+	-	23. Pro/ Arg	**G** **C**,G	G**G**C/ C**C**G	½+	½+
11 STOP /Leu	C	/UAA			24. Ser/ Gly	C,**G** **C**	AGG/ UCC	½+	½+
12 Asp/ Ile	**C** **G**	CUG/ GAU	+	-	25. Ser/ STOP	G	AGU/		
13 His/ Met	**C** **G**	GUG/ CAU	+	-	26. Ser/ Arg	**G** **C**	A**G**C/ U**C**G	+	+
					27. Ala/ Ser	G N	CGG/ GCU	-	-
Total:			7	-				5.5	4.5

Prob.(second-base complementarity)[4]: $p(x \geq 12.5) = 0.65$ NS
Prob. (dual complementarity, 3'-NGN/5'-NCN column): $p(x \geq 4.5) = 0.31$ NS

[1]Based on consensus sequence for tRNA species in the last common ancestor, identified following analysis of 1100 sequences obtained in a search of the Bayreuth database (http://www.uni-bayreuth.de/departments/biochemie/tRNA/). Adapted from results of a previous investigation. [2]

[2]Pairs 4-6, 13, 17-19, 27 contain anticodons with a G:U wobble pair. Formation of pairs with a canonical 36A:34U base pair was hindered by the low frequency of anticodons containing a 5'-A (34-A) in pre-divergence tRNA sequences.

[3]dual compl., refers to tRNA species showing dual complementarity: acceptor-stem second bases (G,C) complementary in a tRNA pair, with complementary anticodons, and with *cis*-anticodon mid-base (35-N) and second base (2-N) identical. Bold letters highlight bases at both sites in these tRNA pairs.

[4]Second base and dual complementarity had non-significant binomial probability. Complementarity among G, C bases at site 2:71 had an intrinsic probability, $v = ½$ per tRNA pair. Dual tRNA complementarity based on complementary G, C bases at site 2:71 and identical bases at *cis* site-2 and -35, had probability, $v = ½ \times ½ = ¼$ per tRNA pair in the 3'-NGN/5'-NCN column.

Second base complementarity in tRNA pairs, with complementary anticodons, can accompany identity with the *cis*-anticodon mid-base. [1] This dual complementarity occurs in six pairs (6/13) of the 3'-NGN/5'-NCN set (Table 1). 2-Base G/C ambiguity in three tRNA pairs (15, 23, 24) reduces the number to 4.5 pairs. Dual complementarity among these thirteen tRNA pairs has a non-significant binomial probability, $p(x \geq 4.5) = 0.31$ NS. Equating semi-matched with full-matched tRNA pairs fails to raise dual complementarity to statistical significance, $p(x \geq 6) = 0.08$ NS, where a one-in-twelve chance exists that the resulting frequency could arise randomly.

Analysis of complementarity in LCA tRNA, whose base sequence was inferred from 1100 tRNA in the Bayreuth database, [4] has refuted, as before, [3] the 2-base and dual complementarity hypothesis. [1] Acceptor stem identity elements, underlying aminoacyl-tRNA-synthetase recognition, according to this, did not shape the genetic code. Involvement of bifunctional cofactor/adaptor tRNA in coordinating code evolution with the growth of amino acid synthesis pathways [2], furthermore, diminishes the role of synthetases in early protein synthesis. Contrary to the '2x2 code' and related 'operational code' hypothesis, [1, 5] a single proto-synthetase evidently attached an amino acid precursor, such as aspartate, to multiple tRNA species, cognate with different sets of codons, prior to the homotopic synthesis of each product amino acid on a designated tRNA cofactor.

Acknowledgement

Dr Sergei Rodin kindly informed me of the 2-base discrepancy in three of the tRNA species examined.

References

[1] Rodin S; Rodin A. (2006). Origin of the genetic code: first aminoacyl-tRNA synthetses could replace isofunctional ribozymes when only the second base of codons was established. *DNA Cell Biol 25*, 365-375.

[2] Davis BK. (2008). Imprint of tRNA diversification on the genetic code: domains of contiguous codons read by related adaptors for sibling amino acids. In, *Messenger RNA Research Perspectives*. Takeyama, T. Editor. New York: Nova Science, pp. 35-79.

[3] Davis, B.K. (2008). Comments on the search for the source of the genetic code. *Messenger RNA Research Perspectives*. Takeyama, T. Editor. New York: Nova Science, pp. 1-8.

[4] Sprinzl, M; Horn, C; Brown, M; Ioudovitch, A; Steinberg, S. (1998) Compilation of tRNA sequences and sequences of tRNA genes. *Nuc. Acids Res. 26*, 148-153.

[5] Schimmel P; Giege R; Moras D; Yokoyama S. (1993). An operational RNA code for amino acids and possible relationship to genetic code. *Proc Natl Acad Sci USA 90*, 8763-8768.